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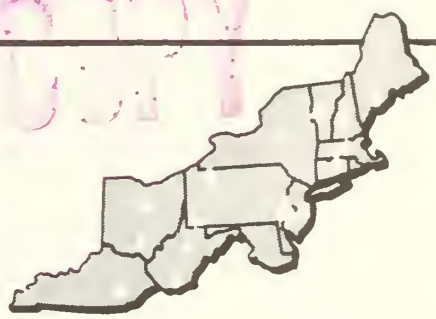

FOREST SERVICE, U.S. DEPT. OF AGRICULTURE, 6816 MARKET STREET, UPPER DARBY, PA. 19082

RADIOGRAPHING PUPARIA OF TACHINID PARASITES OF THE GYPSY MOTH, AND APPLICATION IN PARASITE-RELEASE PROGRAMS

Abstract.—A radiographic technique has been developed for observing and quantifying development and mortality of *Blepharipa scutellata* (Robineau-Desvoidy), *Parasetigena agilis* (Robineau-Desvoidy), and *Compsilura concinnata* (Meigen), tachinid parasites of the gypsy moth, *Porthetria dispar* (L.). Puparia can be examined and sorted immediately after collection and decisions on further collecting and release can be made in the fall. Healthy tachinid pupae can be placed directly in a release area eliminating the handling of adults the following spring.

Recent increase in the geographical spread of the gypsy moth, *Porthetria dispar* (L.), has stimulated new efforts by state and federal agencies to develop biological control programs that can be used effectively, at new points of infestation, to inhibit population increase and prevent further spread. A major component of this approach has been the introduction of parasites into the incipient gypsy moth populations.

Blepharipa scutellata (Robineau-Desvoidy), *Parasetigena agilis* (Robineau-Desvoidy), and *Compsilura concinnata* (Meigen), are three important tachinid parasites of the gypsy moth in natural populations in the Northeast. Procedures for collecting, holding, and releasing these dipterous parasites have changed very little since the first parasite-introduction programs were initiated in the early 1900s (*Burgess and Crossman 1929*).

The general procedure has been to mass-collect gypsy moth larvae and pupae and hold them until the parasite maggot emerges. *C. compsilura* has two to three generations per year. The first-generation maggots are collected as they drop from third- and fourth-instar larvae and then are held in a moist regime until the adult emerges from the puparium. Adults may then be released at desired locations in the same year. *B. scutellata* and *P. agilis*, which have only one generation per year, are allowed to drop directly into sand or some other overwintering medium, or are collected after pupariation (*Fraenkel and Bhaskaran 1973*) and placed in the overwintering site. The next spring the relatively fragile adult flies are collected as they emerge and are released at the desired points.

Although the procedures are relatively simple, a number of practical problems have appeared that greatly reduce the efficiency of the method. 1. Recent investigations into the population dynamics of tachinids at our

laboratory indicate that a high prehibernation mortality of 30 to 40 percent may occur between pupariation and pupal formation. 2. In areas in which the tachinids are collected, the parasites are themselves parasitized, while in the gypsy moth, by the hyperparasite *Brachymeria compsilurae* (Crawford), which may then overwinter as a larva in the tachinid puparium. 3. There is evidence that excessive handling of puparia and adults can cause significant mortality.

Techniques have not been developed for periodically sampling overwintering tachinid puparia to determine the number of healthy, diseased, and parasitized fly pupae. Thus it is currently impossible to predict how many adult flies will be available for release in the present procedures for collecting, holding, and releasing.

The problem of observing and measuring the development and activity of insects when they are concealed in a host or by pupal coverings has been successfully solved for a variety of insects and host material by using radiography (*Holling 1968*; *Demars 1963*; *Graham et al 1964*; *Kirkpatrick and Wilbur 1965*). In 1973, a radiographic technique was developed at the U.S. Forest Service's Northeastern Area State and Private Forestry field laboratory at Stroudsburg, Pennsylvania, for observing and quantifying development and mortality of tachinids while they are concealed in the puparium.

Radiographs of puparia were taken with a Faxitron shielded cabinet X-ray system, model 8050-010. (Mention of brand-name materials should not be construed as endorsement by the U. S. Department of Agriculture or the Forest Service.) A series of test exposures of puparia of various sizes were made, using Kodak Type M X-ray film. The radiographs with the best definition were shot at

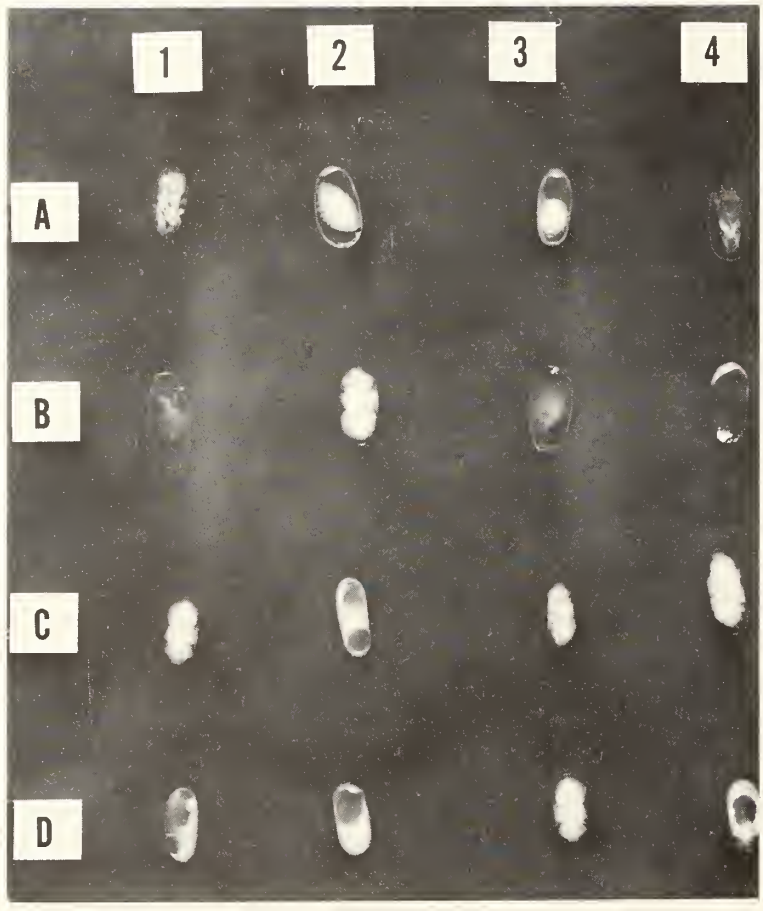

Figure 1.—Radiograph of *Blepharipa scutellata* puparia. Puparia of *Paradetigena agilis* had similar contents and can be identified just as easily.

A1. Hyperparasite, polyembryonic hymenoptera.
A2. Hyperparasite, diapausing hymenoptera larva.
A3. Hyperparasite, diapausing hymenoptera larva.
A4. Dried *Blepharipa scutellata* pre-pupa.
B1. Dried, did not pupate.
B2. Fully formed *B. scutellata* pupa.
B3. Liquified, partially dried.
B4. Empty except for small amount of dried material at posterior end.
C1. Fully formed *B. scutellata* pupa, dorso-ventral view; on a radiograph you cannot distinguish dorsal or ventral because specimen is radio-transparent.
C2. Liquified, two air spaces.
C3. Fully former *B. scutellata* pupa, lateral view.
C4. Fully formed *B. scutellata* pupa, lateral view.
D1. Liquified, partially dried.
D2. Liquified, one large air space at anterior end.
D3. Fully formed pupa, dorso-ventral view.
D4. Liquified, air space in middle.

Figure 2.—Radiograph of *Compsilura concinnata* puparia. All but number 4 contain diapausing hymenopterous hyperparasites. Number 4 was completely dry, but there was no external evidence to indicate that it was empty.

25 KV, 9 mas (large puparia, fig. 1) and 20 KV, 3 mas (small puparia, fig. 2).

Thirty-seven puparia were radiographed in three groups: 10, 10, and 17. (Puparia were held in moist sand or vermiculite until they were radiographed and dissected.) The contents of each puparium were described on the basis of radiographic evidence and then were dissected for verification. Groups were examined, described, and dissected in sequence so that radiographic diagnosis would improve with each group. Puparia were categorized as containing one of the following: (1) a developed pupa; (2) liquified or dried tissue; (3) a hyperparasite. Overall, only two were misidentified and both of these were in the first group (table 1).

Although the technique was developed with a relatively small number of puparia, only a few modifications would be necessary to screen larger collections. Holling (*1958*) developed a lucite holder that permitted X-raying 250 *Neodiprion sertifer* (Geoffroy) cocoons at one time. His technique proved feasible for classifying cocoons containing healthy, parasitized, and diseased sawflies.

Beyond our immediate research requirements, the radiographic technique would be applicable in parasite release programs in several ways:

1. The number of parasites released for either the establishment of the species or as an inundative procedure for immediate control will most certainly require that certain

Table 1.—*Verification, by dissection, of radiographic predictions made for contents of* Blepharipa scutellata, Parasetigena agilis, *and* Compsilura concinnata *puparia.*

State	Radiograph prediction	Dissection
Normal fly pupa	17	19
Liquified or granular	15	14
Hyperparasitized	3	3
Diseased pupa	1	1
Deformed pupa	1	0
	37	37

numbers be available. If mass collection is to be part of such a program, then we must be able to assess the number of healthy pupae in any particular collection. In terms of costs and benefits (man-hours spent collecting), knowledge of the presence of healthy specimens might indicate whether collecting in any particular site would be worthwhile.

2. Fall introduction would eliminate holding cages and the handling of emerging adults in the spring.

3. Fall collections of puparia could be placed immediately in a target area because the possibilities of introducing hyperparasites concealed in puparia would be eliminated.

4. Radiography provides a sampling tool for assessing the overwintering population in any particular area.

Literature Cited

Burgess, A. F., and S. S. Crossman.
1929. Important insect enemies of the gypsy moth and the brown-tail moth. U. S. Dep. Agr. Tech. Bull. 36. 147 p.

Demars, C. J., Jr.
1963. A comparison of radiograph analysis and bark dissection in estimating numbers of western pine beetle. Can. Entomol. 95:1112-1116.

Fraenkel, G., and G. Bhaskaran.
1973. Pupariation and pupation in cyclorrhapous flies (diptera): terminology and interpretation. Ann. Entomol. Soc. Am. 66:418-422.

Graham, H. M., O. T. Robertson, and D. F. Martin.
1964. Radiographic detection of pink bollworm larvae in cottonseed. J. Econ. Entomol. 57: 419-420.

Holling, C. S.
1958. A radiographic technique to identify healthy, parasitized and diseased sawfly prepupae within cocoons. Can. Entomol. 90:59-61.

Kirkpatrick, R. L., and D. A. Wilbur.
1965. The development and habits of the granary weevil *Sitophilus Granarius* within the kernel of wheat. J. Econ. Entomol. 58:979-985.

—T. M. ODELL
P. A. GODWIN
W. B. WHITE

T. M. ODell and P. A. Godwin, Research Entomologists, USDA Forest Service, Northeastern Forest Experiment Station, Insect and Disease Laboratory, Hamden, Conn. W. B. White, Program Manager for Development and Application, USDA Forest Service, Northeastern Area State and Private Forestry, NEFES, Insect and Disease Laboratory, Hamden, Conn.

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